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Prenatal amphetamine and postnatal challenge: The effects on play in rats

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Abstract

The present study examines both the effects of prenatal amphetamine with the effects of an acute amphetamine challenge on play behavior in rats. It was hypothesized that prenatal amphetamine administration would disrupt play behavior in juvenile rats, possibly via a similar mechanism as prenatal cocaine exposure (Wood et al., 1995). It was further hypothesized that the disruptions of play caused by a challenge dose of amphetamine (Beatty, Costello, and Berry, 1984) would differ between rats with prenatal exposure and those without. Our data revealed a significant decrease in play due to postnatal exposure, but no prenatal effect or interaction was observed.

Background: Prenatal Amphetamine Exposure and Social Play

- Exposure to any neuroactive drug during brain development is likely to have far-reaching consequences. The initial structural disruptions continue to influence brain development after birth and the cessation of drug exposure (Mayes et al., 2003).
- Rat models of prenatal exposure to neuroactive drugs such as cocaine have shown significant alterations in social behavior and stress responsiveness (Wood et al., 1994), similar to behavioral alterations seen in human children (Beckwith et al., 1994).
- Prenatal amphetamine exposure in rats results in teratogenic alterations to the norepinephrine (NE) system, including decreased activity of the locus coeruleus and increased NE concentration in the prefrontal cortex (Nasif, Cuadra, and Ramirez, 1999). Notably, prenatal cocaine exposure also seems to alter the NE system, resulting in an increased sensitivity to the NE α-2 receptor antagonist idazoxan (Bayer et al., 2002).
- It is thought that juvenile animals engage in play as a rehearsal for adult behaviors such as aggression or reproductive strategies (Vanderschuren et al, 1997). Social play is often composed of complex yet specific behaviors which are affected by the manipulation of several neurotransmitter systems, including serotonin, dopamine, and norepinepherine (Beatty, Costello, and Berry, 1984; Pellis and Pellis, 1998; Vanderschuren et al, 1997; previous unpublished work in McFarlane lab).
- Although the effects of prenatal cocaine exposure on rat play behavior have been experimentally examined (Wood et al., 1994), the behavioral consequences of exposure to d-amphetamine have yet to be elucidated

Methods

- Pregnancy was determined by detection of vaginal plug or the presence of spermatazoa in a vaginal smear, which was performed daily.
- On the 8th day of pregnancy (pn8) dams began receiving daily drug treatment injected SC between 0800 and 1000 h. Prenatal exposure dams received 4mg/kg/day of d-amphetamine dissolved in 85% saline, and control dams received a comparable dose of saline alone (after Nasif et al., 1999; amphetamine obtained from Sigma, St. Louis, MO).
- Pups from amphetamine litters were cross-fostered to additional control dams within 72 hours of parturition to prevent maternal neglect.
- Pups were weaned on postnatal day 21 (pd21), housed with a same-sex conspecific and randomly assigned to a postnatal treatment group (see **Treatments**).
- Between pd21 and pd30 (peak window for play behavior) each pair underwent behavioral testing consisting of four hours of social isolation (in keeping with lab protocol) followed by a single injection of acute amphetamine (0.5mg/kg in 85% saline) or a similar volume of saline alone.
- 30 minutes after injection, the pair was reunited in an isolated recording chamber and videotaped for a 15-minute trial.
- Two condition-blind raters independently watched each trial and counted the incidence of three selected social play behaviors (see **Scoring Play Behaviors**).

Treatments

		Postnatal (acute) treatment	
		Challenge Amphetamine, 0.5mk/kg	85% Saline
Prenatal Treatment	Amphetamine, 4mg/kg	AA n=12 (6 pairs)	AS n=16 (8 pairs)
	85%Saline	SA n=12 (6 pairs)	SS n=16 (8 pairs)

Scoring Play Behavior

Following/Sniffing: One subject places its nose underneath the tail of the other subject and either sniffs or follows the animal around the cage in the same position.

Boxing/Wrestling: Wrestling involves substantial physical contact between two subjects. Boxing occurs when a subject rears and attempts to make contact with the other subject using its forelimbs.

Pinning: One subject assumes a submissive position (either on its back or side) underneath the other subject.

Statistical Analyses

Interrater reliability was assessed for all three behavioral measures using Pearson's Correlation

Following/ Sniffing: Due to an extremely low correlation score (.19), data from one rater was dropped from analysis. The second rater's scores were used alone after comparison with a third rater (whose scores were incomplete) exhibited a correlation of .776

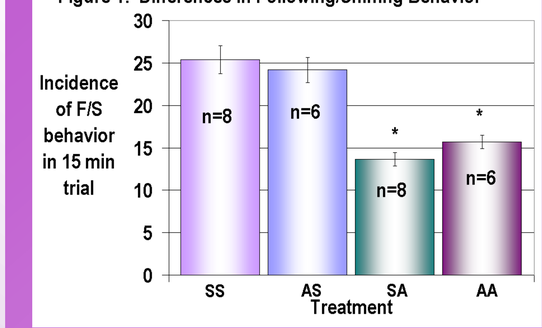
Boxing/Wrestling: Correlation of scores was .922

Pinning: Correlation of scores was .978

Behavior scores for each measure were assessed with 2 x 2 Univariate ANOVA tests, $p<.05$ (see **Figures**)

Results and Figures

Figure 1: Differences in Following/Sniffing Behavior



Following/ Sniffing (Fig 1):
Postnatal amphetamine exposure (groups SA and AA) caused a significant DECREASE in behavior, $F(3,24)=4.937$, $p>.05$ compared to non-prenatally-exposed animals (SA and SS). No significant effects of prenatal exposure (AS and AA, exposed, in comparison with SA and SS, nonexposed) or interaction effects were observed.

Boxing/Wrestling (Fig 2):
Postnatal amphetamine exposure (groups SA and AA) caused a significant DECREASE in behavior, $F(3,24)=45.058$, $p>.05$ compared to non-prenatally-exposed animals (SA and SS). No significant effects of prenatal exposure (AS and AA, exposed, in comparision with SA and SS, nonexposed) or interaction effects were observed.

Figure 2: Differences in Boxing/Wrestling Behavior

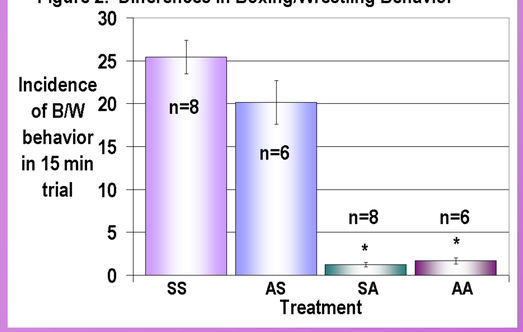
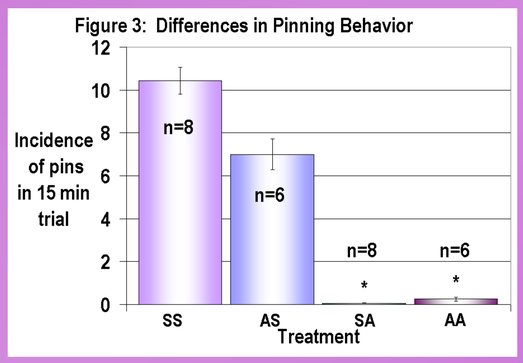


Figure 3: Differences in Pinning Behavior



Pinning (Fig 3):
Postnatal amphetamine exposure (groups SA and AA) caused a significant DECREASE in behavior, $F(3,24)=15.562$, $p>.05$ compared to non-prenatally-exposed groups (SA and SS). No significant effects of prenatal exposure (AS and AA, exposed, in comparison with SA and SS, nonexposed) or interaction effects were observed.

Discussion

The disruption of play behavior by acute amphetamine is consistent with previous literature (Beatty, Costello, and Berry, 1982; Sutton and Raskin, 1985). It is possible that the play-specific behavioral measures of the present study are not sensitive to the behavioral changes in prenatal amphetamine exposure. It is also possible that our dose of prenatal amphetamine was not sufficient to produce any behavioral change; the 4mg/kg/day dose was selected to avoid gross teratogenic effects while still permanently altering brain monoamines (after Nasif, Cuadra, and Ramirez, 1999), but some behavioral studies use daily doses as high as 10mg/kg(Tan, 2003). Finally, the technique of cross-fostering prenatally drug-exposed pups may reduce the behavioral effects of exposure. It has recently been demonstrated that human children of drug-abusing mothers under nonparental care show similar cognitive and social-emotional development to nonexposed children of similar socioeconomic background (Brown et al., 2004).

Useful future experiments would potentially utilize higher doses of prenatal amphetamine or compare cross-fostered pups to pups reared by amphetamine-treated dams. It also remains to be seen whether the current prenatal dose did in fact cause permanent alterations of neurotransmitter systems which are thought to influence play when acutely manipulated, such as norepinepherine and dopamine. If so, the current results indicate that play is a robust behavior. They may point to compensatory mechanisms active during development to act against such systemic disruptions and preserve play, and highlight the importance of social learning components of normal play behavior.

References

- Beatty, W.W., K.B. Costello, and S.L. Berry. (1984) *Suppression of play fighting by amphetamine: effects of catecholamine antagonists, agonists and synthesis inhibitors*. Pharmacology Biochemistry & Behavior, 20: 747-755.
- Bayer, L.E., S. Kakumanu, C.F. Mactutus, R.M. Booze, and B. J. Strupp. (2002) *Prenatal Cocaine exposure alters sensitivity to the effects of idazoxan in a distraction task*. Behavioural Brain Research, 133: 185-196.
- Beckwith, L., C. Rodning, D. Norris, L. Phillipsen, P. Khandabi, and J. Howard. (1994) *Spontaneous play in two-year-olds born to substance abusing mothers*. Infant Mental Health Journal, 15: 189-202.
- Brown, J.V., R. Bakeman, C.D. Coles, K.A. Platzman, and M.E. Lynch (2004) *Prenatal cocaine exposure: a comparision of 2-year-old children in parental and nonparental care.* Child Development, 75: 1282-1295.
- Carlson, Neil R. (2001) *Physiology of Behavior* (8th ed.) (p 589). Boston: Allyn and Bacon.
- Mayes, L.C., D. Cicchetti, S. Acharyya, H. Zhang. (2003) *Developmental trajectories of cocaine-and-other-drug-exposed children and non-cocaine-exposed children*. Journal of Developmental and Behavioral Pediatrics, 24: 323-335.
- Nasif, F.J., G.R. Cuadra, and O.A. Ramirez. (1999) *Permanent alteration of central noradrenergic system by prenatally administered amphetamine*. Developmental Brain Research, 112: 181-188.
- Pellis, S.M., and V.C. Pellis. (1998) *Play fighting of rats in comparative perspective: a schema for neurobehavioral analyses*. Neuroscience and Biobehavioral Reviews, 23: 87-101.
- Sutton, M.E., L.A. Raskin. (1986) *A behavioral analysis of the effects of amphetamine on play and locomotor activity in the post-weaning rat*. Pharmacology, Biochemistry, & Behavior, 24: 455-461.
- Tan, S.-E. (2003) *Prenatal amphetamine exposure alters behavioral reactivity to amphetamine in rats*. Neurotoxicology and Teratology, 25: 579-585.
- Vanderschuren, L.J.M.J., R.J.M. Niesink, and J.M. Van Ree. (1997) *The neurobiology of social play behavior in rats*. Neuroscience and Biobehavioral Reviews, 21: 309-326.
- Wood, R.D., V.A. Molina, J.M. Wagner, and L.P. Spear. (1995) *Play behavior and stress responsivity in periadolescent offspring exposed prenatally to cocaine*. Pharmacology Biochemistry &Behavior, 52: 367-374.

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